8252

LEISHMANIASIS:

A REVIEW OF RECENT LITERATURE.

WELLCOME INSTITUTE LIBRARY
Coll. welTROmec
Call No. Pam WC715 1922 W48e

BY

C. M. WENYON

(*Sectional Editor, Tropical Diseases Bureau*).

[Reprinted from the *Tropical Diseases Bulletin*. 1922. Vol. 19, No. 1, pp. 1–18, and No. 3, pp. 182–193.]

LONDON:
TROPICAL DISEASES BUREAU,
23, Endsleigh Gardens, N.W.1

22200146933

LEISHMANIASIS: A REVIEW OF RECENT LITERATURE.

By C. M. WENYON,

Sectional Editor, Tropical Diseases Bureau.

I. KALA AZAR.

During the past ten years very few important advances have been made in our knowledge of kala azar, with the fortunate exception of the discovery of the specific action of antimony, chiefly in the form of tartar emetic, in the treatment of the disease. We are no nearer the solution of the problem of its transmission and not many new facts of importance have been elicited. No additions to our knowledge have been made, as regards symptomatology and pathology, beyond what may be found in any up-to-date text-book, and no fresh information regarding the structure of the organism causing the disease and its development has come to light. The disease still occurs in the well-known endemic centres of North China, India, the Caspian region, Mediterranean littoral, and the Anglo-Egyptian Sudan, while new foci in Mesopotamia, Sumatra, and Siam, and in the districts around Lake Chad have been discovered. The similar and associated disease which occurs in dogs in the Caspian and Mediterranean regions, and also near Lake Chad, still requires an explanation in view of its entire absence from other endemic centres. Experimental work with animals has attracted considerable attention, but the uncertainty of producing infections has hampered the advances which this line of research was expected to yield. It has been claimed by some observers that many insect flagellates, which have been regarded as peculiar to these animals, are inoculable to vertebrates, and that infections are produced which bear some resemblance to the natural diseases of man and dogs due to leishmania. These observations would tend to confirm the view that leishmaniasis is due to an insect flagellate having invaded the vertebrate. Doubt has, however, recently been thrown on many of these experiments.

Distribution.

Assam still appears to be the most important endemic centre of the disease, and McCombie YOUNG (1920) believes that there is a most disquieting increase in its incidence which calls for immediate action. KÜLZ (1916) reports having found leishmania in the spleen of

(6249) Wt. P13/74 52 5/22 Harrow G75 A

a sick Arab in the Bagdad district of Mesopotamia, and that KADIR, who continued his investigations, had diagnosed kala azar by spleen puncture in eight of twelve cases examined. LOW (1919) treated a case of kala azar in England. According to him the infection was undoubtedly contracted in Bagdad, though LEDINGHAM (1919) considered it possible this had occurred in India five or six months before. The unsatisfactory accounts of the discovery of leishmania in cases of supposed kala azar in the East Indies by DE RAADT (1909), ELDERS (1909) and NEEB (1909) rendered it extremely doubtful if the disease occurred at all in the countries directly east of India. SMITS (1916) has, however, discovered *L. donovani* in six cases locally infected in Sumatra, while MENDELSON (1919) found the parasite in one case from Siam. In China the disease still appears to be limited to the district between the Yangtse and Pekin, as first described by COCHRAN (1913–14). WYLIE (1920) gives an account of 35 cases, 15 of which were diagnosed by spleen puncture, from this region.

In the Mediterranean area the distribution appears to be the whole of the littoral, along which individual centres are constantly being incriminated. Thus, PATANE (1917) has discovered a case in Libya; KLIPPEL and MONIER-VINARD (1921) have recorded the first case from Morocco, and CARNOT and LIBERT (1921) one from Macedonia. MAYER and REINHARD (1918) recorded an infection which was apparently acquired in Asia Minor. The disease, though generally occurring near the coast, may occur further inland, as in Spain. A case was recorded from Lisbon by ALVAREZ in 1910. More recently LABBÉ, TARGHETTA and AMEUILLE (1918) described two cases from Nice in Serbian refugees. They have assumed that the disease was contracted in France. GIOSEFI (1918) observed a case in Trieste. There is no fresh information about the disease in Egypt, nor in the endemic area in the Sudan bordering on Abyssinia, except for records of isolated cases since the issue of the report of the Sudan Kala-azar Commission (1911) and no fresh cases have been recorded from the districts about the Caspian Sea since GURKO's paper in 1913. In 1916 BOUILLIEZ described the first case in a child in the Lake Chad district of West Africa, while TOURNIER (1920) recorded one from the Gaboon. As regards America, the only observation is still that of MIGONE (1913), who saw a case in an Italian who had resided in Paraguay for sixteen years. It is doubtful where he contracted the disease, for he had apparently visited Italy from time to time.

Canine kala azar occurs, as does the human disease, in the Mediterranean littoral and in the Caspian region. As is well known, in certain cases dogs and children have been found infected in the same house. This close association is far from being always observed. NICOLLE (1921) finds that in the Tunis district the canine disease is more widespread than that of children. DELANOË and DENIS (1916) discovered the disease in dogs in Morocco, and it is only recently that the first human case has been seen there. PRINGAULT (1914) found that dogs were infected in Marseilles, but no human cases have been noted in this locality. NELIGAN (1913) found a generalized leishmania infection of a dog in Teheran, but kala azar in man is unknown in Persia. In West Africa the canine disease was noted by LAFONT and HECKENROTH (1915) at Dakar in Senegal. No one has yet discovered kala azar of dogs in India, nor has it been seen in America. Even if we admit that the canine disease is due to the same organism as that producing kala azar in man, it is evident that the dog does not play any essential rôle in its spread, though there is every possibility that it occasionally passes from dog to man or vice versa.

Diagnosis.

The diagnosis of kala azar depends upon the discovery of the parasite, and spleen puncture still remains the most reliable and rapid means of attaining this object. The importance of establishing the diagnosis, especially where new drugs are being tested, is well illustrated by the observation of NICOLLE (1921) in his account of the cases observed in Tunis. Spleen puncture was performed in 172 cases which were suspicious and in only 59 was a diagnosis of kala azar established. Various other methods of discovering the parasite have been advocated, and if these give positive results puncture of the spleen can be dispensed with.

1. *Direct examination of the peripheral blood.*—Many observers, including CHRISTOPHERS, DONOVAN, and especially PATTON, have demonstrated the frequency with which leishmania can be discovered in the peripheral blood of cases of kala azar. PATTON (1912) recorded the results obtained from the examination of 84 cases in Madras; in 42 of these the parasites were discovered in the first film examined, in 13 in the second, in 12 in the third, in 5 in the fourth, in 2 in the fifth, and in 4 in the sixth; in the remaining cases the parasites were not discovered till a larger number of films were examined. Similar results were also obtained by KORKE (1914) in Madras. MACKIE (1914 and 1915), from the examination of 245 cases, found the parasites in the peripheral blood of 21 per cent. of the indigenous Assamese and in 64 per cent. of the tea-garden coolies. More recently KNOWLES (1920) succeeded in 32 out of 72 cases; this result necessitated the examination of 671 blood films. In the Sudan MARSHALL (1912) observed the parasites in the blood of 80 per cent. of cases examined. In the Mediterranean disease NICOLLE and CASSUTO (1908) had observed the parasites in the blood of one case. CANNATA, in Italy (1913 and 1914), discovered leishmania in the blood of 15 out of 16 cases, an observation which required the methodical examination of 20 to 30 films from each case. Similar results were obtained by VAGLIO (1914) in 11 cases, and by RUTELLI (1914) in 7 cases.

It is evident from these results that parasites are frequently present in the peripheral blood. They are generally within mononuclear leucocytes, but may occur in other cells. Their discovery, however, appears to be not always an easy matter, and it evidently necessitates the careful examination of a series of well-prepared films from each case.

2. *Culture of peripheral blood.*—A reliable and easy method of cultivating the organism having been discovered by NICOLLE (NNN medium), it was hoped that blood culture would yield more uniform results than direct examination of the blood. MAYER and WERNER (1914) and WENYON (1914) were the first to demonstrate that cultures could be obtained by inoculating tubes of NNN medium with a few drops of the peripheral blood. ROW (1914), in India, was also successful with a more elaborate method; the blood from the finger was mixed with a citrate saline solution and allowed to deposit for 12 to 24 hours; the deposit was then inoculated into culture media. It is stated that this method obviates the addition to the medium of excess of human serum, which is detrimental to the growth of the parasite. GIUGNI (1914) and CANNATA and CARONIA (1914) also obtained successful results in Italy. CORNWALL and LA FRENAIS (1916) obtained cultures from the blood in seven cases in India. KNOWLES (1920), working in Assam, found that this culture method,

though sometimes yielding very good results, was disappointing as a reliable diagnostic procedure.

Blood culture is certainly a method which should be employed, especially in cases in which spleen puncture is contraindicated. The disadvantage is that the number of parasites inoculated to the medium is so small that often two or three weeks must elapse before flagellates are detectable.

3. *Examination of lymphatic glands.*—COCHRAN (1913) advocated the excision of a superficial lymphatic gland under a local anaesthetic. The gland is cut in half and a smear made from the cut surface. Puncture of the gland without excision did not give good results.

4. *Serum tests.*—The fact that blood serum from kala azar cases was liable to produce a copious precipitate when mixed with water was noted by BRAHMACHARI (1917). In a later publication (1920) this observer advises the addition to the serum of two parts of distilled water. A precipitate was obtained in 20 consecutive cases of kala azar and not in other diseases. RAY (1921) again describes what is undoubtedly the same reaction. To two drops of blood in a tube are added 20 drops of distilled water. In kala azar the mixture shows a turbidity and, on standing, a flocculent precipitate. It seems probable that this explains an observation made by HILL (1913) in China, who noted that in making blood counts in eight consecutive cases of kala azar it was impossible to get the blood to mix uniformly in the pipette, as the blood ran together into clumps. He described the phenomenon as a new sign in the disease. The diluting fluid was a mixture of 1 part of a methyl alcohol solution of Wright's stain with 3 parts of 0·1 solution of sodium chloride in distilled water. It seems that this peculiarity of the blood in kala azar is worthy of further investigation.

Another test which must still be regarded as in the experimental stage is the "formol-gel" reaction advocated by GATÉ and PAPACOSTOS for syphilis. The addition of a couple of drops of commercial formol to about 1 cc. of clear serum causes the mixture to jellify in 24 to 30 hours. SPACKMAN (1921) called attention to the fact that in kala azar the solidification occurred in a few seconds. NAPIER (1921) tested 50 cases of proved kala azar, and noted that solidification of the serum occurred in less than half an hour in 49 of these. In other cases tested, such as malaria, a jelly was not formed till after the expiry of 24 hours. FOX and MACKIE (1921) have devised a simple technique for this reaction. A drop of clear serum is placed on a slide, and this is inverted over a watch glass containing a few drops of formol. In a few minutes the kala azar serum will have solidified, whilst other sera remain fluid. It would appear that this test is of greater promise in kala azar than in syphilis, in which disease it is now regarded as of little, if any, value.

Many observers have attempted to devise a test for kala azar based on the presence of antibodies in the blood. The most extensive investigations are those of CARONIA (1913). He showed that suspensions of the flagellates could be agglutinated or precipitated by the serum of rabbits which had been subjected to inoculations of killed cultures. The serum of naturally infected children showed a reaction which was too feeble to be of diagnostic value, though the administration of a vaccine of killed flagellates increased the potency of their sera, as it did in rabbits. The reaction of deviation of the complement has likewise yielded discordant results.

None of the tests based on serum reactions can be regarded as a substitute for the discovery of the parasite in really scientific work.

Age of Infected Persons.

When it was discovered that kala azar existed in the Mediterranean area, and that here young children were almost exclusively attacked, this form of the disease was named Infantile Kala Azar, and the parasite causing it *Leishmania infantum*, as it was generally considered that in India the disease was essentially one of adults. More recent and careful observations have tended to demonstrate that such a distinction does not actually exist, for in the Mediterranean area adults are not immune to infection, while in India children are more often attacked than was at first supposed. MACKIE (1914) conducted a kala azar survey in the Nowgong district of Assam. The cases were tabulated as Typical, Probable, Suspicious and Doubtful. By performing spleen puncture in a certain number of each group, it was determined that all the typical and probable cases and about half the suspicious ones were actually cases of the disease. Of 195 cases which could thus safely be assumed to have kala azar, 9 were between the ages of 1 and 5, 100 between 6 and 10, 49 between 11 and 15, 17 between 16 and 20, 12 between 20 and 30, and 8 over 30. Of a series of 143 cases treated by MUIR (1917), 25 were under 6 years of age, 56 from 6 to 12, 26 from 13 to 16, 21 from 17 to 25, and 15 about 25. He remarks that kala azar is thus much commoner in children than adults. KNOWLES (1920) gives for 86 patients, all diagnosed by discovery of the parasite, an average age of 21·41 years. Fifty-eight of these cases were between 10 and 26 years of age. There was a maximum age of 40 and a minimum of 2. COCHRAN (1913), in China, found the age in 16 definitely diagnosed cases to vary from 4 to 35 years. In his account of 35 cases, 15 of which were diagnosed by spleen puncture, in the same country, WYLIE (1920) found the ages to vary from 4 to 30 : all of these were males, a fact which is easily understood in view of the known reluctance of the Chinese to allow their women to be seen. In the region of the Caspian it is known that the disease attacks chiefly children and young adults. GURKO (1913) describes four cases from this area. Their ages were $6\frac{1}{2}$, $6\frac{3}{4}$, 16, and 19. In the Sudan, MARSHALL and THOMSON (1911) found that kala azar occurs in persons of 12 years of age and upwards. In the whole of the Mediterranean region the disease is undoubtedly most common in young children, as all the records show. Adults, however, are occasionally attacked. TASHIM (1910) recorded two cases from Tripoli in persons 11 and 18 years of age. GABBI (1909) saw the disease in a youth of 18, a man of 38, and three boys of 13. SPAGNOLIO (1911) saw a case in a man of 35. Similarly, cases in adults have been described by FULCI and BASILE (1911), CHRISTOMANOS (1911), ARCHER (1907), BASSETT-SMITH (1913), TRANTAS (1915), WARD (1916), KERR (1918), CARNOT and LIBERT (1921), and others. It is thus evident that the Mediterranean disease attacks adults and the Indian disease children as in other endemic areas, and it can no longer be maintained that infantile kala azar is distinct from that of India. This conclusion is borne out by all the clinical features of the disease and the character of the organism producing it.

Treatment.

The greatest advance has been made in the treatment of the disease. To GASPAR VIANNA (1913) is due the credit of first bringing to the notice of the medical world the specific action of tartar emetic in cases of South American cutaneous leishmaniasis. His observations

were quickly confirmed, while, in Italy, Di Cristina and Caronia (1915) discovered that the drug was equally efficacious in the treatment of kala azar. Rogers (1915) was the first to employ the drug in India. Since then it has been used on a large scale and has given most satisfactory results. It is now recognized as the accepted method of treatment, and though attempts have been made to discover more easily administered or less toxic substitutes, tartar emetic and its corresponding sodium salt are still the only remedies which are generally employed.

Tartar Emetic. India.—In a letter to the *British Medical Journal* for July 31, 1915, Rogers reported that he had been using tartar emetic in the treatment of kala azar in India with promising results. Accounts of the success of this form of treatment were contained in two papers by Rogers (13 cases) and Muir (9 cases) which appeared in the October number of the *Indian Medical Gazette* for 1915. Of the 13 cases treated by Rogers, 8 were either cured or improved, while 4 either had not changed or were worse, and 1 had died. Muir reported that 8 of the 9 cases treated by him had shown uniform and rapid improvement, while one had died of phthisis. In both series of cases the diagnosis had been established by spleen puncture and discovery of the organism. Rogers and Hume (1916), and Rogers (1916–17) gave details of the treatment of 35 Europeans, 6 of which were children. Diagnosis was made by discovery of the parasite in all of these, except 5 of the children in which the symptoms were sufficiently characteristic to justify the diagnosis of kala azar. Of the 35 cases, 26 were reported as cured, and 8 improved, while one died from phthisis. The cases which were recorded as improved were either still under treatment, or had left hospital prematurely. There had been no known relapse amongst those considered as cured, the majority of which had either been seen or heard of for several months up to a year after discharge from hospital. Rogers (1917) gave the results of the treatment of a series of 13 natives of India, and leishmania was seen in all but one of these. Eight were stated to have been cured, three much improved, and two to have shown no change. In 19 of the cases in which parasites had been discovered and a second puncture of the spleen was performed when cure was considered to have taken place, it was found that leishmania was no longer present. Rogers noted that treatment had to be continued for some time after apparent recovery had taken place, and that the results fully confirmed the sanguine expectations aroused by his previous experience.

After Rogers had introduced the tartar emetic treatment in India it was adopted by other observers. Muir (1916) gave an account of the treatment of a series of about 100 cases, and in 1917 another series of 143 cases, all in natives. In the first series, it is stated that the diagnosis had been made by spleen puncture in the majority of the cases, and, though no details are given, that all cases recover absolutely if treatment is begun before the last stage of the disease is reached. More details are given of the second series of 143 cases. Spleen puncture confirmed the diagnosis in 72 of these, as there was some doubt as to the diagnosis. In the remaining cases the clinical features and blood condition made the diagnosis so clear, that spleen puncture was not performed. Of the 143 cases, all but 46 were discharged as cured. Of the latter, 13 died and 33 discontinued treatment of their own accord, and of these 29 did so because their relatives considered that a cure had already occurred. Muir remarks that it is safe to conclude that of the 143 cases, 126 were either cured or would have been cured if the full course of treatment had been taken.

Another series of 86 cases in which diagnosis was confirmed by finding of the parasite and where tartar emetic treatment was employed is described by KNOWLES (1920). Of the 86 cases, 46 were discharged as cured. In 31 of these, a spleen puncture performed before discharge had failed to reveal any parasites. Of the remaining cases, 11 were progressing favourably towards recovery, while six had absconded before the treatment was completed. One case was evidently dying, and 22 had actually died. The deaths were due to influenzal pneumonia, 6 ; acute dysentery, 5 ; asthma and cardiac failure, 5 ; nephritis and cardiac failure, 2 ; and broncho-pneumonia, dysentery and nephritis, 4. Some of those who died of influenzal pneumonia were already cured of their leishmania infection. Good results following the use of tartar emetic in properly diagnosed cases were reported by MACKIE (1915) one case, BRAHMACHARI (1915) a few cases, GHOSH (1916) four cases and JACKSON (1916) one case, while LOW (1919) recorded the treatment of a case which had apparently contracted the disease in Mesopotamia. Death from influenzal broncho-pneumonia unfortunately occurred when cure had taken place.

In Assam treatment by means of tartar emetic has been adopted on a large scale and records of the success of the treatment are accumulating. In most of these no statements as to the discovery of the parasite is made, so that it is possible that other diseases than kala azar may have been occasionally included. Owing to lack of hospital accommodation and other causes many cases are being treated as out-patients, as mentioned by MUIR (1917). KUNDU (1920) writes of the popularity of the tartar emetic treatment amongst natives in Assam. At the time of writing 45 cases were being treated as out-patients and 16 as in-patients at Nowgong. The treatment of out-patients is unsatisfactory, as the people cease to attend as soon as the initial improvement has taken place, whereas in order to effect a cure treatment must be prolonged.

In 1918 ROGERS recorded the fact that he had completely given up the use of tartar emetic in favour of the corresponding sodium salt—sodium antimonyl tartrate—on account of its lesser toxicity. This salt of antimony had been used by BRAHMACHARI (1916) and GHOSH (1916), with favourable results. ROGERS concluded that it is at least as effective and probably more so than tartar emetic. DODDS-PRICE (1920) has given over 3,000 injections himself, and has watched the effect of twice this number given by his assistants, in Assam. In most cases tartar emetic was apparently used, but he calls attention to the superiority of the sodium salt. He concludes that in these salts we possess the means of curing the vast majority of cases of kala azar, provided treatment is commenced in the early stages of the disease. ELWES (1920) also writes of the good results of the tartar emetic treatment, but agrees that the sodium salt is to be preferred.

It is evident from the above accounts that by the use of tartar emetic or the corresponding sodium salt there is every prospect of bringing about a cure in Indian kala azar, provided the treatment is commenced early enough and is continued for a sufficient time after improvement has taken place. Since its first employment much has been learnt as to the best method of giving the drug and the length of time it is necessary to continue the treatment. In 1917 ROGERS pointed out that the maximum dose required to destroy the parasites, when given every third day, or twice a week, over two or three months, is about 1 cc. of a 2 per cent. solution for every 10 lb. of body-weight. The maximum dose for a European, however, must not exceed 10 cc. and for an Indian

7 cc. This maximum dose must only be reached gradually after commencing, in persons of 12 years of age and upwards, with about 5 cc. if there is no debility. In children and old people the initial dose must not exceed one-third of 1 cc. per 10 lb. of weight. Cases were defined as cured when the fever had completely ceased for two or more months, when there had been considerable gain in body-weight, and a restoration of the blood, and especially of the white corpuscles, to the normal, and a decided diminution in the size of the spleen.

Attention was drawn to the toxic effects of tartar emetic. It was realized that great care had to be exercised in administering the drug, and that it was necessary to commence with small doses and gradually to increase them. Three deaths following injections of tartar emetic are mentioned, but in these cases it was found that the solution contained a white precipitate. ROGERS (1919), however, recorded a case which died as a result of the toxic action of the sodium salt. The necessity of using only perfectly clear solutions is emphasized.

MUIR (1916–17) stated that injections should be given every second day till fever had been absent for a month and then once a week for two to four months. Unless these weekly injections are continued relapse may occur.

Some very interesting facts were elucidated by KNOWLES (1920) during his experiences in Shillong with tartar emetic. The first effect of treatment is the immediate disappearance of parasites from the peripheral blood. The second result is their gradual disappearance from the spleen, while the third is the cessation of fever. At the same time there is a steady increase in body-weight, decrease in size of the spleen, and a return of the blood picture to the normal. The rate of disappearance of parasites from the spleen varies, but, speaking generally, they may be said to become scanty and difficult to find after 100 cgm. have been given, while after 200 cgm. a cure is assured. That the parasites are still alive after considerable doses have been given is proved by the fact that cultures were obtained after the administration of 18, 31, 49, 89 and even 174 cgm. The spleen steadily diminishes in size and may even become impalpable. This latter result is not always seen, for the spleen may still be palpable even in cases which have evidently been sterilized of their infection. During treatment the liver may actually increase in size, and it is suggested that this may be a result of accumulation of tartar emetic in the tissues. The directions given by KNOWLES (1920) for the administration of tartar emetic as practised at Shillong are as follows :—

" (*a*) The potassium salt is the one used at Shillong ; and the ' heavy powder ' should alone be used ; not the ' light powder.'

" (*b*) The solution is of strength 1 per cent. in normal saline ; autoclaved at 110° C. for ten minutes, and kept in a rubber-capped flask.

" (*c*) If there be the least trace of deposit or opalescence in the solution it must be discarded, and is not safe to use.

" (*d*) Beginning with an intravenous dose of 3–4 c.c. for an adult, the injections are given on alternate days, and the dose gradually raised to 10–12 c.c., above which dose it is not advisable to go. At first the full effect of the injection seems to fall upon the parasites ; later, as the patient becomes cleared of infection, the tissues seem to become *more* susceptible to the drug. There is little, if any, evidence of acquired immunity to antimony, and there may be, to some extent, accumulation in the liver. Hence, towards the end of treatment the dose should *not* be pushed and the intervals between doses should be increased.

" (*e*) The standard course at Shillong is a course of 200 cg., distributed over a period of two and a half to three months. .

" (*f*) The injections should not be given within two hours of a meal.

" (*g*) Accessory treatment by tonics and the clearing of any helminthic infection is essential. Sodium morrhuate is useful in fattening patients.

" (*h*) In the presence of complications the tartar emetic should be continued, unless there are very serious contra-indications. In a recent case, where a child appeared for ten days to be *in extremis* with what seemed to be the terminal dysentery, the injections were continued and the child made a good recovery at the time. . . . Nephritis is the most difficult complication to treat, but as the nephritis is the result of interstitial fibrosis of the kidney from parasite infection, the injections should be continued cautiously. In brief, complications are an indication to reduce the dose and proceed cautiously, but do not necessitate the abandonment of treatment.

" (*i*) The state of the heart and the examination of the urine should be attended to. The sudden appearance of oedema may be a danger signal. In giving the injections at Shillong, as also in doing spleen puncture, pituitrin and adrenalin are kept handy, although there is hardly ever any occasion to use either drug.

" (*j*) No accidents, sepsis, thrombosis, phlebitis or other ill effects have been seen. The use of a rather large needle (20 c.c. Roux syringe needle) seems better for intravenous work than that of a very fine needle, as it is easier with the former to make certain of being in the lumen of the vein. Count 'eight' slowly over the injection of each c.c. The 2 per cent. solution—to judge from one or two cases treated with it before arrival at Shillong—seems more liable to cause local irritation if the vein be missed. This is not seen with the 1 per cent. solution."

Mediterranean area.—As already pointed out, tartar emetic for the treatment of kala azar was originated by Di Cristina and Caronia (1915) in Italy. In their first paper they describe the treatment of 10 cases, the ages of which varied from 15 months to 6 years. The solution used was a 1 per cent. tartar emetic, which was injected with a fine needle into the median basilic, external jugular, or temporal vein. The commencing dose was 2 cgm., and this was increased to 10 cgm. Of the cases cured the eldest child received 84, and the youngest 35 cgm. Two cases which were in the final stage of the disease died soon after treatment was commenced, while a third died of acute nephritis. The remaining 7 cases were either cured or were reported as progressing so favourably that a cure was ensured.

Jemma (1916) announced that he had obtained a cure in 21 out of 26 cases in Naples, while Spagnolio (1916) recorded the successful treatment of 4 cases. The most extensive series of Italian cases consisted of 110, which were treated at Jemma's clinic in Naples, between October, 1913, and July, 1920, an account of which has been given by Javarone (1920) and Foti and Javarone (1921). This series evidently includes the 26 cases originally reported by Jemma. In all a diagnosis had been made by spleen puncture. The most noticeable feature of this series is the difference in the mortality-rate before and after the introduction of the tartar emetic treatment—whereas recovery had been the exception, it now became the rule. Of the 110 cases, 63 were treated by tartar emetic, with 49 recoveries and 14 deaths. A 1 per cent. solution of the drug was used, and was run into the vein from a graduated burette. For children aged 3 the first dose is 2 cc., and this is increased to 3, 4 and 5 cc. In older children the dose is 2 to 3 cc., increased to 6 or 7 cc. At least two clear days must elapse between each injection. The treatment must be continued for 40 or 50 days, during which 30–50 cgm. of the drug is given. The deaths were due in all cases to intercurrent maladies either during or after treatment. Some of those who died

were actually cured of the leishmania infection. As was to be expected, the mortality-rate was highest amongst the youngest children.

An attempt to treat 43 children in an out-patient department was described by LONGO (1917). The results were far from satisfactory owing to the unwillingness of the parents to submit their children to the prolonged course of treatment.

TREATMENT OF KALA AZAR BY TARTAR EMETIC.

Observer.	No. of Cases.	Diagnosed by discovery of parasite.	Cured.*	Improved (treatment in progress).	Spleen puncture negative after treatment.	No improvement, or treatment not completed.	Deaths.	Death, but leishmania infection apparently eradicated.
			INDIAN CASES.					
ROGERS, 1915	13	13	8	—	—	4	1	—
MUIR, 1915..	9	9	—	8	—	—	1	—
ROGERS & HUME, 1916	6	6	5	—	6	—	1	1
ROGERS, 1916	12	11	8	4	5	—	—	—
ROGERS, 1917	17	12	15	2	—	—	—	—
ROGERS, 1917	13	12	8	3	8	1	—	—
MUIR, 1917..	143	72	97	—	—	33	13	—
KNOWLES, 1920.. ..	86	86	46	11	31	7	22	2
MACKIE, 1915	1	1	1	—	—	—	—	—
GHOSH, 1916	4	4	4	—	—	—	—	—
JACKSON, 1916	1	1	1	—	—	—	—	—
LOW, 1919 ..	1	1	—	—	—	—	1	1
			MEDITERRANEAN CASES.					
DI CRISTINA & CARONIA, 1915.. ..	10	10	5	2	—	—	3	—
JEMMA, 1916	26	26	21	—	—	—	5	—
SPAGNOLIO, 1916.. ..	4	4	4	—	—	—	—	—
FOTI & JAVARONE, 1921	63	49	—	—	—	—	14	8

* The data are insufficient to admit of a precise statement of the period of observation after treatment.

It is quite clear that tartar emetic is just as specific in the treatment of the kala azar of the Mediterranean area as it is in India. The difficulties of carrying out continued intravenous medication in young children make it all the more desirable that some simpler method of administering antimony should be discovered. Furthermore, it has been shown that in some cases death has resulted from administration of tartar emetic. Some individuals react badly to the drug, so that it is necessary to commence with small doses and gradually work up to the maximum, a careful watch being kept on the patient after each injection. Shortly after a dose has been given a cough very frequently is developed. In other cases there may be definite rigors, rise of temperature, vomiting, and diarrhoea. Some patients complain of rheumatic pains and a general feeling of depression. It is most important, however, when once the course has been commenced to continue it till recovery has taken place, as pointed out by ROGERS, MUIR, KNOWLES, and others.

Other compounds of antimony.—Though it is generally admitted that the majority of cases of kala azar can be permanently cured by means of tartar emetic, the fact that injections of this drug are not entirely free from unpleasant consequences has led to the trial of other antimony derivatives.

Paraacetyl amidophenyl stibinate of sodium.—The introduction of this salt, which is known by the name *Stibacetin,* in the treatment of kala azar is due to CARONIA (1916). The drug, which was first prepared in Germany, is easily soluble in water, and contains about 33 per cent. of antimony.* The solution must be prepared fresh. It was tried by intramuscular injection in four cases in Italy. For children under two years of age an initial intramuscular injection of 3 cgm. was given, and was increased to 10 cgm. In older children the dose ranged from 5 to 15 cgm. The injections can be given every second day into each buttock alternately. Of the four cases a boy aged $3\frac{1}{2}$ years had completely recovered after three months' treatment, during which he received 3·6 gm. of the drug; a girl aged 23 months was cured by 9 injections in the course of 16 days, and another aged 12 months by 6 injections in 10 days; in the latter case an abscess formed after an injection of 12·5 cgm.; the fourth case, a boy of 17 months, improved considerably, but died from an abscess in the lumbar region, which was present before treatment was commenced. The treatment of a single case, with favourable result, was reported by KHÀRINA-MARINUCCI (1916). Under the name of *Stibenyl,* which is said to be a closely allied compound, but which is undoubtedly *Stibacetin*, BRAHMACHARI (1921) reports the use of this drug in two cases. In one it was given intravenously, in the other intramuscularly. The doses varied from 0·1 to 0·3 gm. It is not known if a permanent cure was effected. MANSON-BAHR (1920 and 1921) had good results in one case by the intravenous method, while in another a dose of 0·4 gm. produced vomiting and diarrhoea, the patient ultimately dying of amoebic dysentery. Leishmania, proved by culture to be living, were still present in the organs at post-mortem examination. In spite of reports he has received of alarming symptoms following the use of this drug (undoubtedly an impure form of *Stibacetin*) in India, he still believes that this compound holds out better prospects for the treatment of kala azar than tartar emetic or the sodium salt. The experience on which this statement is made is, however, not very extensive.

FOTI and JAVARONE (1921), in Italy, used *Stibacetin* in 10 cases. Eight were treated by intramuscular injections and two intravenously. For intramuscular injections they commenced with a dose of 3 cgm. in

---

* The following information has been kindly supplied by Dr. T. A. HENRY, of the Wellcome Chemical Research laboratories :—

Stibacetin—Sodium paraacetylaminophenylstibinate, $C_{24}H_{25}O_{10}N_3Sb_3Na$, contains theoretically 40·12 per cent. of antimony. The form of *Stibacetin* sold as *Stibenyl* in this country in May 1920 contained 34·85 per cent. of antimony, and that now on sale, which is advertised as Von Heyden's, contains 33·16 per cent. Two different batches contained 32·54 and 33·79 = mean 33·16. Sodium antimonyl tartrate, $C_4H_4O_7SbNa,\frac{1}{2}H_2O$, contains 38·06 per cent. of antimony, and the commercial salt as made for use in medicine is pure. Potassium antimonyl tartrate, $C_4H_4O_7SbK,\frac{1}{2}H_2O$, contains 36·14 per cent. of antimony, and the commercial salt is pure.

All the foregoing percentages are expressed as percentages of metallic antimony for the formulae as given, including water of crystallization where shown in the formulae.

the form of a solution (2 cgm. in 1 cc. of cold distilled water). The injections are given at intervals of two days, and the dose is increased to 10 cgm. The duration of treatment is longer than when the intravenous method is employed, but this disadvantage is compensated for by the ease of administration. The drug must not be given subcutaneously, as it causes necrosis. Of the 10 cases treated seven were cured and three died.

The number of cases treated with *Stibacetin*, all of which were properly diagnosed, cannot compare with the large number in which tartar emetic has been successfully employed, but it is evident that the drug has a similar action; and if it can be administered intramuscularly, we have at any rate a method of treatment which is more suitable for the handling of the disease in young children, in whom intravenous injections are difficult to carry out. So far, however, there is no evidence that it is more potent than tartar emetic.

Colloid antimony sulphide.—BRAHMACHARI (1917) stated that he had tried colloidal sulphide of antimony, both intramuscularly and intravenously, in the treatment of kala azar. As the results were disappointing he discontinued its use. ROGERS (1919) gave a more detailed account of the treatment of 10 cases of undoubted kala azar by means of this drug. The method of preparation is fully described. Doses up to 20 cc. of a solution slightly weaker than 1 in 500 were given repeatedly by the intravenous route without the least toxic symptoms. The results of this treatment were excellent, and the 10 cases, which received from 8 to 37 cgm. of the drug, were all cured. The advantages of this drug over tartar emetic are said to be its lower toxicity, the more rapid gain in weight of the patient, and the smaller quantity required to effect a cure. ROGERS regards it as a distinct improvement on the soluble antimony tartrates in the treatment of kala azar.

Other compounds of antimony.—BRAHMACHARI, who has experimented with a large number of antimony preparations, reports good results following the intravenous use of metallic antimony in suspension and colloidal metallic antimony. Of the former preparation he says it is the strongest preparation of antimony for the treatment of kala azar, being superior to any others that were tried before its use. Another drug used by this observer is hyper-acid antimonyl tartrate (+ urethane). The highest dose was 5 cc. of a 2 per cent. solution given intramuscularly. The use of this strength was not free from pain. A 1 per cent. solution, however, is almost painless. Of this drug he writes that it is one of the greatest advances in the treatment of kala azar. This investigator (1915–17) also reported the treatment of two cases of kala azar by intravenous injections of formaldehyde. The drug was said to act like a specific, but, curiously enough, he states that he has not been able to give it a further trial though we know from his publications that he has been conducting further investigations on the treatment of kala azar with other antimony derivatives, of which, in 1921, he has given an account. None of them has given results superior to those obtained with tartar emetic. It is evident, therefore, that though he has reported so very favourably on some of the drugs he has used, he is not satisfied with his own results, as he is still passing on to new compounds of antimony. In no instance reported has any of these drugs been used in a large enough series of cases to justify the deductions made as to their value; and it is to be regretted that before further experimentation with new derivatives, those so favourably judged have not been fully tested.

Animal Experiments.

One of the great hindrances to the satisfactory study of the method of spread of kala azar is the lack of an animal which is susceptible to the disease. It may safely be affirmed that if strains of leishmania were as readily inoculable to animals as are the pathogenic trypanosomes, our knowledge of the etiology of the disease would be considerably more advanced than it is at present. Both the Indian and Mediterranean viruses are inoculable to dogs, monkeys, rats and mice, but the infections are produced only after inoculation of large doses of virus, and then with no degree of certainty. When infection takes place it is of a chronic nature, and though parasites are sometimes numerous in the organs, at other times they are very scanty and difficult to detect. Recent investigations have done nothing in the way of solving this problem. It results, therefore, that it is very difficult to test the infectivity of any invertebrate host which may be suspected of transmitting the disease. Dogs and monkeys appear to be the most susceptible animals, and they have been infected with the virus obtained both from man and from naturally infected dogs. It is possible, by inoculating larger doses of virus from animal to animal, to maintain any particular strain. LAVERAN (1920) has shown that in the passage of a virus through mice the virulence gradually decreased. Mice, in some instances, were found to be infected two years after inoculation. Rats appear to be more difficult to infect than mice, and guinea-pigs and rabbits still more so. There is apparently no difference in the infections produced in animals by the Indian and Mediterranean viruses.

Interesting experiments with the virus of kala azar were made by ROW (1912–13). By inoculating a monkey in the skin of the head by scarification with virus from the spleen of a case of kala azar, nodules were produced after an incubation of three to five months. In these leishmania were present and there was no sign of a general infection. Similar lesions were produced in another monkey by the subcutaneous injection of cultures. The material from one of these nodules injected into the skin reproduced the local lesion only, while a general infection was produced in those monkeys injected intraperitoneally. It thus appears that *Leishmania donovani* produces local cutaneous lesions or a general infection according to the method of inoculation. KORKE (1914) repeated ROW's experiments, with similar results. He found, however, that in some cases in which the local skin lesion had resulted from cutaneous inoculation there existed also a generalized infection, while in others only a generalized infection developed after inoculation in the skin. Infections are more readily produced in animals by injecting the virus from a man or other animal than by cultures of the organism.

Transmission.

The problem of the transmission of kala azar still requires solution. In the Mediterranean area it is supposed by BASILE that the flea is the vector, but it must be admitted that no further proof of this is forthcoming. This observer claimed to have followed the development of *Leishmania donovani* in the flea, and to have successfully transmitted the disease from infected to healthy dogs by means of these insects. Several other observers recorded successful experiments of the same kind. On the other hand, certain investigators (DA SILVA, 1913; WENYON, 1914) failed entirely to obtain any

development in fleas and were unable to effect a transmission of the disease. The fact that fleas are very liable to have a flagellate infection of their own renders it difficult to trace the development of leishmania in these insects. Furthermore, it is not an easy matter to exclude an infection in a living dog, so that in the apparently positive transmission experiments it is always possible that the supposed healthy dogs had already a latent infection. In such experiments, in which it may take several months for an infection to reveal itself in the animals employed, the exclusion of other sources of infection during the experiment is almost impossible. In a recent paper BASILE (1920) reviews the present position of this subject and restates his views. He claims to be able to distinguish the natural flagellates of the flea from those which develop from the leishmania ingested. Moreover, he states that the developing leishmania eventually produce cysts in the hind gut of the flea, as does the natural flea flagellate, and that kala azar may be acquired not only by the flea inoculating leishmania at the time of biting, but by an individual ingesting the encysted forms passed in the faeces of the flea. These and other statements made by this author are highly speculative, and lack the necessary experimental proof. He brings forward no convincing evidence in favour of the flea transmission hypothesis, and no facts which would exclude the fallacies of his earlier experiments.

On the transmission of kala azar in India recent investigations have thrown little light. Numerous insects have been investigated, and of these the bed bug has claimed the most attention. It is well known that PATTON first demonstrated that in the stomach of the bug leishmania are able to develop to the flagellate stage and undergo a multiplication. It was claimed by other observers that this development did not prove that the bug was the true host, and that the development took place probably because the stomach of the bug was acting as a culture tube. This was borne out by the fact that other flagellates are able to undergo a temporary multiplication and development in the bug's stomach. CORNWALL and LA FRENAIS (1916) showed that the flagellate forms of *L. donovani* could survive for at least 29 days in the intestine of the bug. In a recent paper PATTON and RAO (1921) have investigated the persistence of leishmania in the gut of the bug by making cultures on NNN medium at intervals after feeding. From a bug which had fed on cultures of *L. donovani*, and had had no subsequent feed, a culture was obtained from the mid-gut on the thirty-first day, while from another bug a culture from the hind gut was made on the thirty-fourth day. A culture was obtained from the mid-gut of a bug which had fed on cultures 41 days before and had had subsequent feeds of blood. It is thus evident that the parasite of kala azar can survive for at least 41 days in the gut of the bug, and this fact might suggest that the bug is the host of the parasite. That this is not necessarily the case is proved by the experiments of PATTON, LA FRENAIS and RAO (1921), who found that *Leptomonas pulicis* and *Crithidia ctenocephali* of the flea and *Herpetomonas muscae domesticae* could also survive for many days in the bug, the latter flagellate for at least 45 days. The forms of the parasite which occur in the bug are similar to those which are found in cultures in NNN medium. CORNWALL and LA FRENAIS (1916) described a certain form of the parasite which occurred as the "*thick-tailed*" *flagellate*. It is a rounded body, 5μ to 6μ in diameter, and has a long flagellum four or five times as thick as that possessed by the ordinary form.

Further observations on the behaviour of *L. donovani* in bed bugs were made by Helen ADIE (1921) in India. This observer fed bugs on spleen pulp from cases of kala azar. The appearances recorded occurred in certain bugs which had died after feeding, and had been left in saline solution in an incubator for a few days before being examined. In these bugs it was found that an active multiplication of the flagellates had taken place, and that this had occurred not only in the cavity of the gut, but also in the cells lining it. It is supposed that the flagellate, probably the thick-tailed form of CORNWALL and LA FRENAIS, enters the cell and becomes eventually a large multinucleated sphere, which segments into a number of daughter forms. This intracellular development is compared with the intracellular development of *Trypanosoma lewisi* which occurs in the cells of the flea's gut. It is unfortunate that satisfactory evidence of a similar development was not obtained in living healthy bugs, though large numbers were examined. It is impossible to resist the conclusion that this development was merely another form of culture which had taken place in the dead and degenerate cells. The figures purporting to depict the various stages of this development are far from satisfactory, so much so that in many cases it is impossible to recognize the bodies as leishmania at all. Those which are intended to show the multiplication of leishmania by schizogony can equally well be interpreted in another manner. They might even be stages of schizogony of some sporozoon ; and that other parasites might be present is borne out by the fact that nematode larvae are also depicted as occurring in these bugs. It may safely be accepted, therefore, that the bed bug hypothesis of the transmission of kala azar is still unproved.

Several observers claim to have observed leishmania in the faecal mucus of kala azar cases. KNOWLES (1920) examined mucus from such cases, and though he saw bodies having a certain resemblance to leishmania they were so atypical that he was quite unconvinced as to their leishmanial nature. The examination of ankylostomes and their larvae hatched from eggs from kala azar cases by this observer failed to throw any light on the problem. The method of transmission of kala azar is still a mystery. ARCHIBALD (1914), who succeeded in infecting a monkey by feeding it on kala azar spleen material has suggested that infection may be carried in water, but there is no evidence of this. It is possible, as WENYON (1914) suggested, that the parasite of kala azar is an insect flagellate which normally passes directly from one insect to another, as do most of the purely insect forms, and that occasionally man is infected. If such were the case it would be unnecessary to suppose that the insect invariably infects itself from the vertebrate host, though it might do so. CORNWALL and MENON (1918) suggested the possibility of kala azar being conveyed by the rupture of an infected bug on the excoriated skin.

Relation of Insect Flagellates to Leishmaniasis.

In 1913 LAVERAN and FRANCHINI published an account of certain experiments they had conducted with *Leptomonas ctenocephali* of the dog flea, *Crithidia fasciculata* of *Anopheles maculipennis*, and *Crithidia melophagi* of the sheep ked. It was claimed that rats and mice could be infected with these flagellates by intraperitoneal inoculation. Furthermore, they stated that mice which were kept with infected fleas or had been fed on these insects became infected. Similar experiments were reported by FANTHAM and PORTER (1914–16) in

the case of dogs, mice, birds, fish and amphibia. These investigations promised to throw a new light on the relationship of insect flagellates to vertebrates, and suggested that kala azar or oriental sore might be the result of infection of man with flagellates which had hitherto been regarded as peculiar to the invertebrates. LAVERAN and FRANCHINI, in a series of papers, extended their observations to other flagellates, including those of the house fly, sand fly (*Phlebotomus*), and even Euphorbia plants. These observers claim that the inoculated animals develop an infection which bears a striking resemblance to those due to leishmania, in that leishmania forms of the parasite occur in the blood, spleen and liver, or other organs. Flagellate leptomonas forms also were found in the blood. These infections are said to be transferable by subinoculation to other animals, which in some cases were killed by the presence of the organisms. Many of the statements made by these authors are difficult to understand. The apparent ease with which the infections were produced in their experiments stands in marked contrast to the uniform failure of other observers. Furthermore, the experiments with the flagellate of the sheep ked, which is almost certainly the invertebrate stage of the sheep trypanosome, is described as producing an infection of the kala azar type rather than one which would be expected to arise from the inoculation of a trypanosome. In this connection the recent investigations of HOARE (1921) are of significance. This observer attempted to repeat some of the experiments above described. Though these were conducted with abundant material and with the greatest care, no single instance of an infection of a vertebrate by an invertebrate flagellate was demonstrable. In the light of these results the uniform and rapid successes above described are very difficult to explain, and it is doubtful whether they throw any light on the problem of the transmission of kala azar. The experiments of previous observers have been collected in tabular form by HOARE (1921), and readers who are interested in the details are referred to his paper.

Culture of Leishmania.

It is now known that *L. donovani* can be easily cultivated in the water of condensation which forms in the tubes of Nicolle's rabbit-blood-agar medium first described by MANCEAUX (1911), and generally known as the Novy, McNeal, Nicolle (NNN) medium. It was also shown by NICOLLE and MANCEAUX (1911) that growth took place on the surface of the solid medium above the water of condensation. This medium has now been extensively used, and it has been abundantly proved that strains of leishmania can be maintained indefinitely by subcultures in it. NICOLLE (1921) has stated that a strain of *L. donovani* has been thus grown for over ten years at Tunis, and that during this period it has been subcultured over 291 times.

THOMSON J. G. and SINTON (1921) record the successful culture of *L. donovani* from the bone marrow of a case of kala azar 14 hours after death. The case, which has been referred to by MANSON-BAHR, had received a course of injections of tartar emetic. Successful cultures have, however, been previously obtained at post-mortems as late after death as 36 hours.

Prophylaxis.

As we have no definite knowledge of the etiology of kala azar it is evident that any prophylactic measures must, of necessity, be of an empirical nature. As BASILE and other Italian observers believe

that the canine disease of the Mediterranean area is identical with that occurring in children and that the flea is the agent of transmission, the removal of dogs from close association with human beings and the destruction of possibly infected dogs have been advocated as a means of prevention. BASILE (1916) records that in Bordonero in Sicily, where both the human and canine diseases were common, most of the infected dogs were destroyed. In the following year there were no fresh cases in children, and in the year after only one, though in that in which the dogs were killed there had been seven new cases in a population of 2,000.

In India prophylactic measures have been based on the known fact that kala azar is a house and family disease. KNOWLES (1920) gives some very instructive instances, in which several members of one family were found suffering from the disease. The following groups are recorded :—Mother and nine brothers ; father, sister and three brothers ; wife, daughter, sister and one brother ; mother and two brothers ; two brothers and sister-in-law.

In 1914, DODDS PRICE and ROGERS published an account of the uniform success of segregation measures in eradicating kala azar from Assam Tea Gardens. On one estate new coolie lines were erected 300 yards distant from the old lines. Of 240 persons, 96 who had no cases of kala azar in their families were removed to the new lines. The new lines were filled up with new coolies to the extent of 800. This transference was carried out in 1897 and sixteen years later the new lines were still free from the disease, while the coolies in the old lines had died during the following few years. Kala azar, furthermore, spread from the old lines to a contiguous line, in which 60 healthy coolies were housed. One-third of these died during the next fifteen months and all the rest succumbed or left the estate during the few succeeding years. Other illustrative examples are quoted in the same paper in support of house infection as an important factor in the spread of the disease.

MCCOMBIE YOUNG (1920) describes the measures which are being adopted in Assam to cope with the spread of kala azar. Known endemic areas are annually inspected, and when a village shows a large number of cases the disease is considered as hyper-endemic and the population is removed to a new locality,the definitely infected, the contacts, and the uninfected individuals being segregated separately. In the case of areas which show infections for the first time more radical measures are adopted. The population of every village which shows even a single case is removed to a new site. It is too early to speak of the results of these radical measures.

MCCOMBIE YOUNG points out that the newly introduced treatment by tartar emetic promises to facilitate the eradication of the disease. It is becoming widely known, and its success appreciated by the native population.

* REFERENCES.

ADIE (1921), *Indian Jl. Med. Research*, ix, 255.
ARCHER (1907), *Jl. Roy. Army Med. Corps*, ix, 287–290, 511–513.
ARCHIBALD (1914), v, 265.
BASILE (1916), viii, 8.
—— (1920), xvii, 367.
BASSETT-SMITH (1913), iii, 133.
BOUILLIEZ (1916), viii, 2.
BRAHMACHARI (1915), viii, 5.
—— (1917), xi, 152.
—— (1917–20), "Kala Azar and its Treatment" (1st and 2nd editions). Calcutta: Butterworth & Co. (India).
—— (1921), xviii, 239.
CANNATA (1913), ii, 436.
—— (1914) iii, 545.
—— & CARONIA (1914), iv, 397.
CARNOT & LIBERT (1921), xviii, 239.
CARONIA (1913), iii, 134.
—— (1916) viii, 7.
CHRISTOMANOS (1911), *Deutsch. Med. Woch.*, xxxvii, 641–644.
COCHRAN (1913), iii, 130.
CORNWALL & LA FRENAIS (1916), viii, 403.
—— & MENON (1918), xiii, 98.
DI CRISTINA & CARONIA (1915), v, 269.
DELANOË & DENIS (1916), *Bull. Soc. Path. Exot.*, ix, 202.
DODDS-PRICE (1920), xvi, 454; xvii, 357.
—— & ROGERS (1914), iii, 552.
ELWES (1920), xvii, 357.
FANTHAM & PORTER (1915), v, 280.
FOTI & JAVARONE (1921), xvii, 358.
FOX & MACKIE (1921), *Indian Med. Gaz.*, Oct. 334.
FULCI & BASILE (1911), *Rendic. Accad. Lincei.*, xx, ser. 5, 1 sem., pp. 132–136.
GABBI (1909), *Boll. d. Cliniche.*, Milan, xxvi, 251–261.
—— (1909), *Arch. Inst. Pasteur Tunis*, iv, 209.
GHOSH (1916), viii, 6; ix, 235.
GIOSEFI (1918), xiii, 99.
GIUGNI (1914), iv, 396.
GURKO (1913), ii, 447.
HILL (1913), ii, 447
HOARE (1921), xviii, 12.
JACKSON (1916), ix, 235.
JAVARONE (1920), xvi, 454.
JEMMA (1916), viii, 6.
KERR (1918), xiii, 99.
KHARINA & MARINUCCI (1916), x, 66.
KLIPPEL & MONIER-VINARD (1921), xviii, 238.
KNOWLES (1918), xv, 434.
—— (1920), xvii, 351.
KORKE (1914), v, 273.
KUNDU (1920), xv, 436.
LABBÉ, TARGHETTA & AMEUILLE (1918), xii, 198.
LAFONT & HECKENROTH (1915), vi, 223.
LAVERAN (1920), xvii, 367.
—— & FRANCHINI (1913), iii, 122.
LONGO (1917), xi, 153.
LOW (1919), xiv, 202.
MACKIE (1914), v, 262.
—— (1915), vii, 79.
MANCEAUX (1911), *Bull. Soc. Path. Exot.*, iv, 286–288.
MANSON-BAHR (1920), xvi, 376.
—— (1921), xviii, 240.
MARSHALL (1911), i, 3.
MAYER & WERNER (1914), iii, 136.
MENDELSON (1919), xiv, 238.
MIGONE (1913), i, 635.
MUIR (1915), viii, 3.
—— (1916), ix, 234.
—— (1917), xi, 151.
NAPIER (1921), *Indian Med. Gaz.*, Sept., 238.
NELIGAN (1913), ii, 449.
NICOLLE (1921), xviii, 238.
—— & CASSUTO (1908), *Arch. Inst. Pasteur Tunis*, 3.
—— & MANCEAUX (1911), *Bull. Soc. Path. Exot.*, iv, 712–713.
PATTON (1912), i, 1.
——, LA FRENAIS & RAO (1921), *Indian Jl. Med. Research*, viii, 629.
—— & RAO (1921), *Indian Jl. Med. Research*, ix, 252.
PRINGAULT (1914), iv, 398.
RAY (1921), xvii, 360.
ROGERS (1915), vi, 221.
—— (1915), viii, 3.
—— (1916), ix, 231.
—— (1917), xi, 145.
—— (1918), xii, 199.
—— (1919), xiv, 203
—— & HUME (1916), viii, 3.
ROW (1912), i, 357.
—— (1914), v, 260.
—— (1914), xi, 144.
DA SILVA (1913), iii, 547.
SMITS (1916), viii, 372.
SPACKMAN (1921), *British Med. Jl.*, Aug. 13, 266.
SPAGNOLIO (1911), *Studi Mal. Trop. Ital.*, 44.
—— (1916), x, 65.
TASHIM (1910). *Arch. Inst. Pasteur Tunis*, 138.
TOURNIER (1920), xvi, 454.
TRANTAS (1915), vii, 79.
VAGLIO (1914), iv, 396.
VIANNA (1913), *Arch. Brazil de Med.*, ii, 426.
WARD (1916), viii, 406.
WENYON (1914), iii, 543, 550.
WYLIE (1920), xvii, 356.
YOUNG (McCombie) & SCOTT (1913), vi, 250.
—— (1920), xvii, 355.

* References are to summaries in the *Tropical Diseases Bulletin* unless otherwise stated. Dates in parentheses refer to year of publication of original papers.

LEISHMANIASIS: A REVIEW OF RECENT LITERATURE.

By C. M. WENYON,
Sectional Editor, Tropical Diseases Bureau.

II. ORIENTAL SORE.

The chief advances which have been made in our knowledge of oriental sore fall into three categories. Firstly, there is the gradual accumulation of evidence that species of Phlebotomus may be the carriers of the disease, at least in the Old World; secondly, the wide distribution of leishmania infections in South and Central America, where an exceedingly chronic disease exists, which, though mostly of the cutaneous variety, in 10 to 20 per cent. of the cases extends to the naso-pharynx; thirdly, the great advance made in the treatment of the disease by the discovery of the specific remedy, tartar emetic.

Distribution.

The most remarkable feature of the distribution of oriental sore is the well-known fact that in certain countries the endemic centres are sharply marked off from those of kala azar. In India, oriental sore occurs in the districts north-west of a line joining Cambay and Delhi, while kala azar is found only east of this line. A similar condition maintains in North Africa, where kala azar is found in the districts round Tunis, while oriental sore is found chiefly at Gafsa, about 2 degrees further south. Recent records show that this distribution still maintains. In other areas it appears that such a demarcation does not exist. Both kala azar and oriental sore occur in the districts around the Caspian Sea, VON PETERSEN (1914) having obtained details of 1,200 cases in Turkestan. The same is very largely true of the Mediterranean littoral, and of the Blue Nile districts of the Sudan.

Oriental sore in the Eastern Hemisphere has a wide distribution in the part of India already indicated, Arabia, Persia, Mesopotamia, the Caspian region and, generally speaking, all the countries bordering on the Mediterranean. MARTOGLIO (1912) has recorded the disease from Abyssinia, while WAGON (1914) has seen it in Dahomey, in Nigeria, and BOUILLIEZ (1917) in the French Congo. The disease had previously been noted in the Niger district by STÉVENEL (1911) and BENOIT-GONIN (1911). The most interesting recent observation is that of RAVAUT (1920), who has diagnosed a case in France in a girl who became infected at Beixas in the Eastern Pyrenees. Another case reported by the same observer appears to have originated in Spain, where the disease was first recorded by CAMACHO (1914).

CASTELLANI (1913) reported a case of oro-pharyngeal leishmaniasis in Ceylon, while similar cases have been recorded by CHRISTOPHERSON (1914) and SUSU (1917) from the Blue Nile district of the Sudan. These are of interest in that they exhibit a type of the disease which is common in South America, but exceedingly rare in the Old World, where the lesions are almost exclusively of the cutaneous variety.

Since the discovery by LINDENBERG (1909) and CARINI and PARANHOS (1909) of leishmania in the ulcer of Bauru in Brazil, and the

demonstration by CARINI (1911) of the presence of the parasites in the nasopharyngeal lesions, which are often associated with the cutaneous condition, numerous records of the distribution of the disease in South America have been published. It has a wide distribution throughout Brazil, but the whole of the northern half of South America and Central America contain endemic centres of the disease, which is especially prevalent in the virgin forest regions which are in process of development in connexion with new railway construction. It must not be supposed that involvement of the mucosa of the nose, mouth and pharynx occurs in every case of leishmania infection in South America. In Brazil, according to DA SILVEIRA (1920), this condition occurred in 20 per cent. of 15,000 cases seen at S. Paulo between 1914 and 1919. INCHAUSTEGUI (1918) finds that in certain parts of Mexico amongst the chicle gum gatherers there may be an incidence of dermal leishmaniasis of 50 per cent., the ear being the most common site of the lesion. The disease had previously been reported from Yucatan by SEIDELIN (1912) and RODRÍGUEZ ARJONA (1915). BATES (1913) has, however, recorded a case from Panama, which showed involvement of the nasal mucosa. In the Guianas the disease, which is known as Forest Yaws, Boschyaws or Pian Bois, and was first shown to be due to leishmania by NATTAN-LARRIER, TOUIN and HECKENROTH (1909) and FLU (1911), seems to be limited almost exclusively to the skin. THÉZÉ (1916) pointed out that the mucosae were rarely affected and that spontaneous healing of the cutaneous sores occurred in 2 or 3 years. BONNE, however (1918), has seen cases in Guiana in which the nasal mucosa was involved. ITURBE (1917) first recorded the disease from Venezuela. It has been studied in Colombia by TEJERA (1920), who finds that the mucosae are rarely affected. MIGONE (1913) and LINDSAY (1914) have described the disease from Paraguay ; STRONG and his co-workers (1913) met with cases in Peru, while ESCOMEL (1917) gives an account of the disease as it occurs in Peru and Bolivia.

Natural Disease in Animals.—Though many observers had noted that dogs are liable to an ulcerative condition of the head in endemic centres of oriental sore, it was not till 1913 that NELIGAN demonstrated leishmania in these lesions in dogs in Teheran. In one dog there appeared to be a general infection at the same time. GACHET (1915) observed ulcers in 15 of 21 dogs examined at random in Teheran. Leishmania were found in these lesions. YAKIMOFF and SCHOKHOR (1914) found the parasites in sores on dogs in Tashkent, while PEDROSA (1913) recorded the discovery of leishmania in an ulcer on the nose of a dog in an endemic centre of the human disease in Brazil, the only instance of a natural infection in South America.

Clinical.—Of the few clinical records worthy of note attention may be called to the cases of involvement of the mucosae of the nose and mouth recorded by CASTELLANI (1913) in Ceylon, and by CHRISTOPHERSON (1914, 1917) and SUSU (1917) in the Sudan. In CASTELLANI's case it appears that the disease commenced as an ulceration of the soft palate without any primary skin lesion. It is a remarkable fact that neither oriental sore nor kala azar occur in Ceylon, so that further information about cases of this kind is desirable. The two cases described by CHRISTOPHERSON (1914) and SUSU (1917) resembled the South American form of the disease, as there was involvement of the nasal mucosae which appeared to have arisen from an extension of ulcers on the neighbouring skin. There was also injection of the gums and hard palate. From the

figures produced it would appear that these conditions were actually due to leishmania, though it is remarked that the organisms were exceedingly scanty and difficult to find. It has always to be remembered that under certain conditions, yeasts may simulate leishmania very closely.* That the second Sudan case, first seen by SUSU and subsequently treated by CHRISTOPHERSON (1917) rapidly recovered under tartar emetic, though the disease had been in progress for four years, is a point in favour of its leishmania nature. The photographs of these two Sudan cases are strikingly reminiscent of the numerous illustrations which have appeared of the severe naso-pharyngeal type of the disease in South America.

An unintentional inoculation experiment conducted on himself by BOUILLIEZ (1917) is of interest, in that it gave rise to a type of infection not observed under natural conditions. Fluid from a mouse infected with *L. tropica* was accidentally introduced into the eye. Four months later a small nodule appeared on the conjunctival surface of the lower lid. It increased in size till it reached that of an almond. Numerous leishmania were discovered in the lesion. Cure was brought about by intravenous injections of tartar emetic. It is a well-known fact that it is easy to reproduce the disease in man by inoculation of the skin with virus from an oriental sore.

The cutaneous lesions of the South American disease appear to be more chronic in character than the oriental sore of the East. In some cases there is a tendency for the lymphatic system draining the affected area to become involved. WERNER (1911) drew attention to this condition and demonstrated leishmania in the enlarged glands. Quite recently DE AGUIAR PUPO (1921) has described a case in which swellings appeared along the course of the lymphatics of the leg. These broke down, producing ulcers in which leishmania occurred.

Diagnosis.

As regards the diagnosis of oriental sore, the discovery of the parasite is the only method which gives reliable information. The organism can be found in smears made from material obtained by scraping the granulations, after removal of the scab if such be present. Very frequently, however, the leishmania obtained in this manner are scanty in numbers and in a degenerate state owing to the adverse conditions resulting from secondary bacterial invasion and the resulting pus formation. More normal forms can be obtained, in cases which have commenced to ulcerate, by running a fine glass pipette into the base of the ulcer through its marginal skin. Sometimes it may not be possible to find parasites even in this way. A diagnosis may occasionally be made in such cases by the culture method. This is best carried out by sterilizing the marginal skin of the ulcer with iodine and running in a fine glass pipette through a puncture made in the skin, the object being to get beneath the ulcer so as to obtain material free from bacterial contamination. With the cells in the

* In this connexion it is perhaps worthy of note that HUNTEMÜLLER (1914) described a new parasite from Jericho Boil under the name of *Plasmosoma jerichoense*. The writer was able to examine preparations containing this supposed organism. It was evident that it was merely a badly fixed leishmania. Jericho Boil is now known to be oriental sore. Similarly, CHALMERS and KAMAR (1920) recorded as *Toxoplasma pyrogenes* Castellani, 1913, certain structures which were found post mortem in a case of febrile splenomegaly from the Barun district of the Sudan. This district is known to be an endemic centre of kala azar, and from information the writer has received he is in a position to state that the supposed Toxoplasma was merely a degenerate and altered leishmania.

pipette, N.N.N. medium is inoculated. In heavy infections, flagellates appear in 48 hours, but in very scanty ones three weeks may elapse before they are present in the medium in sufficient numbers to be recognized.

Treatment.

As pointed out in the section on kala azar, intravenous medication with tartar emetic was first introduced by GASPAR VIANNA in the treatment of the dermal and mucosal leishmaniasis of South America. Since then it has been employed in the treatment of oriental sore in the Old World. The number of records of its use do not appear to be as great as might be expected. SCOTT (1917) was the first observer to publish an account of the treatment of oriental sore by this method. He was successful in 63 cases of Frontier Sore in India. Unfortunately, leishmania were found only in 4 of 27 cases examined microscopically. SINTON (1917) cured six diagnosed cases in a period of 8 to 37 days. CHRISTOPHERSON (1917) obtained a good result in a severe case in the Sudan; though the disease had been in progress for four years rapid recovery took place. GREIG (1917) treated 18 Mesopotamia cases, with recovery in 17, in a period varying from 16 to 52 days. BOUILLIEZ (1918) was cured of an accidental infection of the conjunctiva by intravenous tartar emetic after injections of luargol, 606, and neo-salvarsan had failed. LOW (1920) and PATRICK (1920) report the cure of single cases, while SINTON (1921) gives a further account of the treatment of six cases in Turkestan. He remarks that in this district the disease appeared to respond more readily to intravenous tartar emetic than in Mesopotamia, where he had had previous experience. The fact that oriental sore in the Old World is a comparatively benign disease which invariably cures itself usually in 6 to 18 months, and is followed by no serious consequences save the scar, is probably the reason why intravenous injections of tartar emetic have not been employed in its treatment as freely as in kala azar. Large numbers of cases of oriental sore occurred during the war amongst troops in Mesopotamia, and many medical men were reluctant to adopt the intravenous medication with tartar emetic for a disease which was accompanied by no constitutional disturbance and which was known to be self-curing. Thus ORMEROD (1920) says that intravenous tartar emetic is efficacious but appears to be a drastic line of treatment for a localized skin lesion. It also requires detention of the patient in hospital. SINTON (1917), however, writes that the danger of the intravenous tartar emetic treatment seems slight if care is taken not to push the drug to excessive limits and to see that the patient has no kidney trouble before starting a course of treatment. The scarring left after this method seems less than after any other. It gives much less trouble to both physician and patient, besides being very much quicker than the usual means.

GREIG (1917) gave an account of 76 cases which had been sent to Karachi from Mesopotamia. With a view to obtaining comparative data of different methods of treatment, the cases were divided into five groups as follows :—(1) Control cases untreated, 24 ; (2) Intravenous injections of tartar emetic, 18 ; (3) Intravenous tartar emetic together with local inunctions with tartar emetic ointment in strength increasing from 2 to 15 per cent., 14 cases ; (4) Oral administration of tartar emetic together with inunction, 17 cases ; (5) Inunction only, 3 cases. The results, which are somewhat vitiated from the fact that the majority

at least had been subject to previous treatment in Mesopotamia, are set forth in the following table:—

TABLE I.

	Groups.				
	1	2	3	4	5
No. of cases	24	18	14	17	3
Average duration of disease to admission to hospital	2·5 (months)	2·6 (months)	2·8 (months)	2·3 (months)	3 (months)
No. of days cases were under observation in hospital:—					
a. Average	21·9	28·2	32·7	25·1	18·6
b. Maximum ..	54	52	59	59	17
c. Minimum ..	5	16	19	4	15
No. of cc. of 1 per cent. A.T. administered intravenously:—					
a. Average	Nil	71·5	83	Nil	Nil
b. Maximum ..	Nil	150	179	Nil	Nil
c. Minimum ..	Nil	20	22	Nil	Nil
No. of cases:—					
Healed	20	17	13	12	3
Improved	1	0	1	1	0
Not healed	3	1	0	4	0

As the untreated cases only persisted for an average of 21·9 days, it is evident that many of the cases were well on the road to recovery when they came under observation. The method of administering tartar emetic intravenously for oriental sore is the same as that employed for kala azar.

Local Treatment.—The older methods of treatment by excision and the application of escharotics, such as powdered permanganate of potash and carbon dioxide snow, have had their advocates in more recent literature. Though these methods undoubtedly suffice in many cases to bring about a rapid cure, attempts have been made to find a more specific local treatment which will be less drastic.

Treatment by Tartar Emetic Ointment.—LOW (1915) successfully treated a case by the application of a 2 per cent. tartar emetic ointment. Another case was treated by LOW (1920) with this ointment as well as by intravenous injections of tartar emetic. In the table given above, GREIG (1917) records the successful treatment of three cases by means of the ointment; it is admitted, however, that these cases were recovering when they arrived in hospital. In LOW's first case, treatment with salicylic acid ointment and methylene blue ointment for five weeks had only made the sore worse. Tartar emetic ointment was then applied daily for a fortnight, when it was stopped and replaced by a simple dressing. A complete healing had taken place three weeks later. In the second case, tartar emetic injections were

given twice a week for three weeks, but these were discontinued owing to attacks of malaria, and local treatment with ointment was commenced a week later, with the result that the sore had healed in a month.

Treatment by X-rays.—This line of treatment was adopted in Mesopotamia during the war. The best account is given by ORMEROD (1920), who found it to be the most practicable method for the following reasons :—(1) The rays act directly on the causative organism ; (2) They are able to penetrate unbroken skin or masses of diseased tissue, thereby being efficient in ulcerated and non-ulcerated sores alike ; (3) Cases can be treated as out-patients and can very often continue their occupation ; (4) There is no risk of constitutional disturbance, the danger of dermatitis or necrosis being absent with the doses used ; (5) The treatment is short, painless, and easy to administer ; (6) Sores in awkward positions such as the inner canthus of the eye, lips and alae nasi can very readily be dealt with ; (7) The scars left after healing are supple and of such a colour in about six months as to be barely noticeable. Of 84 cases, showing in all 136 sores, the average time from the commencement to complete healing of the sores was 27 days, with a minimum of 10 days and a maximum of two months. The description of the technique is given in full in this *Bulletin*, Vol. 17, p. 364. MITCHELL (1921) states that at a Basra hospital after a trial of every possible method of treatment it was found that a single full pastille dose of X-rays produced a cure in 10 days in the majority of cases. TRESTON (1921) also reports good results from X-ray treatment.

Treatment by Ionization.—EVANS (1918) found this method to give most satisfactory results. The ulcer is cleared of all septic material and crusts and is covered with a pad extending for half an inch beyond the edge of the sore. The pad consists of 16 layers of lint soaked in 2 per cent. zinc sulphate solution and is firmly applied to the ulcer under a zinc electrode by means of a bandage. The pad is connected with the positive pole of the current. The indifferent pad, which must be of much larger size, is placed on any convenient part of the body. An arm or a leg bath with saline solution may replace the indifferent pad. The current is supplied by 18 accumulators, giving an average of 36–38 volts. This is connected with a short resistance so that the current can be gradually increased. A patient with a sore having an area of about one square inch is usually able to stand 10 milli-ampères. The application is continued for 20 minutes, during which the pad is constantly moistened with zinc sulphate solution. Of eight cases cured at the time of writing, the number of applications and the number of days required for the sore to heal were 5–30, 4–22, 5–36, 2–6, 5–8, 3–8, 2–7 and 6–12 respectively. MACCORMAC (1919) treating a case by ionization with sodium hypochlorite obtained rapid healing.

Treatment by Injection of Emetine.—PHOTINOS (1920) described a new method of treatment for oriental sore in Crete, which he had found efficacious in 23 cases. He injected a solution of emetine hydrochloride around and below the sore, both subcutaneously and intracutaneously. For a small sore the total amount of the drug necessary was 0·01 gm., while for large sores he used 0·05 gm. The injections are made with a fine needle and the solution is forced into the skin at various points in the whole area around the sore. In sores which require the above doses only one sitting is necessary,

complete healing taking place after the lapse of 15 to 30 days. In larger sores or multiple lesions it is necessary to inject the requisite quantity of drug at intervals of a few days. CALICETI (1920) treated two cases of Sicilian origin by this method. Two injections were given of 8–10 cgm. of emetine hydrochloride and cure was complete in three weeks.

Other Methods of Treatment.—GRAY (1921) reports that during 1917 and 1918, in Mesopotamia, he treated over 200 cases of oriental sore by scraping and cauterizing with carbolic acid. The average length of time required to bring about a cure was 17 days, and it was his experience that relapse was uncommon and the scars less obvious than after other methods of treatment.

DUCKWORTH (1921) states that he found the following method very efficacious in Mesopotamia. After cleaning the sore a piece of lint the exact size of the ulcer was soaked in a solution of urotropin (1 drachm to the ounce) and applied to the sore. The applications, which are made twice daily, are accompanied by continued healing of the sore. TRESTON (1921), from experience gained in Mesopotamia, advocates the following lines of treatment :—(*a*) For sores in the early stages, X-ray applications and crystals of potassium permanganate ; (*b*) For fairly advanced cases, crystals of potassium permanganate and intravenous tartar emetic ; (*c*) For sores in the advanced stages, thorough scraping under chloroform and application of ol. ricini dressings ; (*d*) For sores on the face, other than those in very early stages, intravenous tartar emetic.

For treatment of the more serious type of the disease met with in South and Central America, GASPAR VIANNA's treatment by intravenous injections of tartar emetic is still mostly employed. There have been many reports of the good results following its use. It is admitted that the drug acts more readily on the purely cutaneous condition, though frequently remarkable recoveries occur in the more serious naso-pharyngeal cases with long histories extending over many years. TORRES (1920) describes the method of treatment in detail. It consists in the injection of 0·06 to 0·07 of a gm. as an initial dose, increasing at each injection by 0·02 gm. up to a dose of 0·12 to 0·14 gm. After fifteen of these injections, a week's rest from treatment may be necessary. At the same time the local skin and pharyngeal or nasal lesions are treated with a 0·1 per cent. solution of tartar emetic. Some cases, however, do not respond to this treatment, in which case "protosan," a preparation made by the Instituto Oswaldo Cruz, has been used with advantage. He describes one case which had 248 separate lesions on different parts of the body. Treatment with tartar emetic and protosan was being carried out with definitely good results when the patient contracted a fatal pneumonia. TEJERA (1920) records the case of an Englishman who had a sore on the ear : 50 injections of 10 cgm. of tartar emetic were given without any but a temporary improvement. ESCOMEL (1917) states that he has successfully treated several cases as out-patients with Martindale's oxide of antimony solution. An injection of 6–8 cc. can be given each week or 1–2 cc. every two days. The drug keeps well and can be given subcutaneously, intramuscularly or intravenously.

It is evident that for some reason not properly understood tartar emetic occasionally fails to effect a cure in the South American disease. This is also true to some extent of the more benign oriental sore of the Old World.

Animal Experiments.

NICOLLE and SICRE (1908) first showed that inoculation of the skin of monkeys either with virus from a human sore or with culture of *Leishmania tropica* was followed by the development of a sore in which leishmania could be demonstrated. NICOLLE and MANCEAUX (1910) proved that dogs were similarly susceptible. Since then, many animals have been successfully inoculated by LAVERAN and others; the outcome of these experiments clearly demonstrates that oriental sore of both the Old and New World can be readily produced in dogs and monkeys, and with less certainty in other animals. GONDER (1913) was the first to show that a general infection in mice could be produced by intraperitoneal or intravenous injection of culture of *L. tropica*. In the infected animals there was great enlargement of the liver and spleen, while there developed on the legs, tail and head, oedematous swellings which finally ulcerated. In all these lesions, leishmania was found. GONDER noted that in infections produced with the virus of infantile kala azar, no skin lesions resulted. It was possible to hand on the infections from mouse to mouse by sub-inoculation. Extensive experiments with mice were conducted by LAVERAN from 1914 onwards. He was able to confirm GONDER's results as regards the production of a general infection and the skin lesions, and further demonstrated that some of the mice acquired a heavy infection of the tunica vaginalis which caused such a thickening of this structure as to give rise to visible tumours. The infection was inoculable from animal to animal, but it was only in certain mice that the heavy infections occurred. In severe infections there was swelling and ulceration of the legs, feet and head, while the lesions sometimes revealed leishmania in enormous numbers. Recovery from the milder infections was the rule, and, as had been previously shown by NICOLLE in the case of monkeys and dogs, the recovered animals were susceptible to reinoculation. Many observers have failed to infect mice, so that it appears that it is only occasionally that a strain is obtained which is pathogenic to these animals. LAVERAN was also successful in infecting guinea-pigs, rats and gerbils.

An exhaustive account of experimental inoculation of both *Leishmania tropica* and *Leishmania donovani* is to be found in LAVERAN's treatise on Leishmaniasis published in 1917.

Transmission.

Oriental Sore in the Old World.—Of the many theories as to the mode of transmission of oriental sore which have been propounded, that which is received with most favour at the present time attributes it to the bite of the sand fly, a view which was first suggested by PRESSAT in 1905. In the same year the brothers SERGENT attempted to produce an infection by the bites of this insect at Biskra. In 1911 the writer suggested the sand fly as a possible vector in Baghdad. In 1915, SERGENT (Ed. & Et.), LEMAIRE and SÉNEVET attempted to produce infection in man, monkey and mouse by the bites of *Phlebotomus minutus*, and also by inoculation of the skin with crushed flies; the results were negative. ACTON (1919) drew attention to the fact that the sites of Phlebotomus bites on the body corresponded with those of oriental sore as it occurred in Mesopotamia, and that the bites of other insects did not thus correspond. In 1911 the writer discovered that 6 per cent. of Phlebotomus in

Aleppo harboured a flagellate of the leptomonas type, an observation which was confirmed by MACKIE (1914) in *Phlebotomus minutus* in India, and by LAVERAN and FRANCHINI (1920) in *P. papatasii* in France. PATTON (1919) stated that both *P. minutus* and *P. papatasii* were known to harbour the flagellate in Mesopotamia, yet in 1922 he writes that *P. papatasii* is infected in Palestine and *probably* in Mesopotamia. Cultures of the flagellates were obtained by LAVERAN and FRANCHINI in N.N.N. medium and with them they claim to have produced a general infection in mice and guinea-pigs, and on the skin of dogs local lesions resembling oriental sore. The incrimination of the sand fly has been taken a stage further by the brothers SERGENT, PARROT, DONATIEN and BÉGUET (1921), who describe the production of an oriental sore on the arm of a human being in Algiers two and a half months after a scarified area of the skin of the arm had been treated with saline solution in which had been crushed seven *Phlebotomus papatasii*, collected at Biskra three to four days before. On August 16 and 17, 1921, a batch of 94 sand flies was collected at the military hospital of Biskra, a well-known endemic centre of oriental sore. They were despatched to Algiers, where the disease is not known, a distance of 600 kilometres. Only seven *P. papatasii* arrived alive. These were broken up in saline solution and the fluid applied to a scarification in the upper third of the forearm of a human being. The wound healed without leaving any trace. Two months and twenty-four days after the experiment a small papule with central incrustation was noted at this spot. It was examined the next day and was found to contain numerous leishmania as in oriental sore. It is interesting to note that in the fluid applied to the sore no flagellates nor other organisms were visible.

This experiment demonstrates that the *P. papatasii* can harbour for at least three days a virus which is able to produce a lesion resembling oriental sore in its characters, in the parasites it contains, and in the length of the incubation period required for its development.

As the observers remark, it is only by further work that it will be possible to determine the origin of the virus in the fly, whether derived from a reservoir or a flagellate infection natural to these flies, and the exact mechanism of infection in nature.

Another aspect of the sand fly problem is the view first enunciated by SERGENT, LEMAIRE and SÉNEVET (1914), that the gecko (*Tarentola mauritanica*) may act as a reservoir for the virus of oriental sore. In support of this view they note that the lizard is ravenously fed upon by sand flies, and, furthermore, that a flagellate of the leptomonas type was culturable from the organs of 15·7 per cent. of the geckos. LAVERAN (1915) attempted without success to infect geckos with *Leishmania tropica*. The presence of the flagellate in geckos was again noted by CHATTON and BLANC (1918), who, moreover, obtained a temporary development in bed bugs fed on geckos, an observation which may throw some light on the behaviour of leishmania in these insects. Later in the same year these observers found that the geckos of Tunis were free from the flagellate found in the animals further south. They accordingly inoculated geckos intra-peritoneally with cultures of *Leishmania tropica*. In one experiment three cultures from the blood were positive in three of twelve geckos which had been inoculated one, twelve and thirteen days before. In another experiment in which the geckos were left at a temperature of 22° and 23° C., three out of four were positive on the twelfth day. NICOLLE, BLANC and LANGERON (1920) repeated these experiments and found that the flagellates occurring in cultures of the natural

gecko flagellate were morphologically distinguishable from those in cultures of *Leishmania tropica.* The blood of a naturally infected gecko was inoculated intradermically to a man on two occasions and also to a monkey, while three men and a monkey were inoculated with cultures of the flagellate. No infection occurred in any case. They also failed to produce an infection in uninfected geckos by inoculating the blood of infected geckos and cultures of the flagellate. They conclude that the flagellate is only accidentally present in the blood of geckos and that it is probably of intestinal origin. LAVERAN and FRANCHINI (1921) found that the blood of geckos of Italy also harboured this flagellate, while FRANCHINI (1921) discovered that a flagellate of the leptomonas type also occurred in the rectum, thus upholding the view of NICOLLE, BLANC and LANGERON, that the blood flagellate was the result of accidental invasion of an intestinal form. On the whole there seems to be little real evidence that the gecko plays the part of reservoir for the virus of oriental sore.

PARROT (1919) records an observation which is of interest in the light of the gecko experiments just described. Eight female sand flies and six geckos were brought in a cage from a place where oriental sore is endemic to another where the disease does not occur. The flies escaped and three months later three individuals in the house developed oriental sore. One of these was the medical man who had collected the flies and had been exposed to infection elsewhere, but the other two had not been in an endemic zone for one, and three and a half years respectively. Even supposing the disease was caused by the escaped flies, there is no proof that they were not already infected when caught.

Bed Bug.—PATTON, who was the first to suggest the bed bug as a vector for oriental sore in Cambay in India, still maintains that this is the carrier so far as Cambay is concerned, though he believes (PATTON, 1919) that in Mesopotamia the sand fly is to blame. His reasons, which have been restated in a recent paper (1922), for suspecting the bed bug are that he considers the parts of the body most usually bitten by bugs are those in which oriental sore most commonly occurs, that the parasite *Leishmania tropica* becomes a flagellate in the stomach of the bug, as was first demonstrated by the writer (1911), and that he has been able to discover development in the stomach cells of bed bugs like that described by Mrs. ADIE (1921) for *Leishmania donovani.* It does not appear, however, that this development was seen in living bugs. CORNWALL and LA FRENAIS (1922) conducted experiments with both *L. tropica* and *L. donovani* with a view to obtaining evidence of an intracellular stage. The bugs were fed on cultures and at varying times after feeding the gut was removed, fixed and sectioned. By this technique the exact relation of parasite to cells is maintained. Though heavy flagellate infections of the lumen of the gut were found, there was no evidence of any intracellular stage, an observation which is in support of the view expressed in Part 1 of this Review (p. 15) that the intracellular stages observed by Mrs. ADIE and by PATTON possibly represented merely a culture in dead or dying cells. CORNWALL and LA FRENAIS also state that the thick-tailed form of the parasite in the bug, first seen by CORNWALL, is a form which has no place in the true cycle of development of the flagellate. There seems to be even less evidence in favour of regarding the bed bug as a carrier of oriental sore than of kala azar. Mrs. ADIE (1922) telegraphs that she has found a swarming infection of Leishman-Donovan bodies in the salivary glands and ducts of a bed bug taken from the bed of a

suspected case of kala azar.* This discovery possibly provides further ground for suspecting the bed bug, but cannot, as she claims, supply the positive proof that kala azar is transmitted by the bite of the bug. There was no evidence that this particular bug had fed upon a kala azar case, nor that the flagellate observed was actually the kala azar parasite, which cannot be distinguished on morphological grounds from many purely insect flagellates occurring not only in the intestine but also in the body cavity, salivary glands and other organs of insects.

House Fly.—BLANC and CAMINOPETROS (1921) conducted an inquiry into the distribution of oriental sore in the Island of Crete. No evidence of any vertebrate reservoir of the disease could be obtained and they concluded that man is the only host, and that the disease is handed on from one to another by direct contact. Flies were allowed to feed on sores and five hours later the legs and heads were crushed in a drop of liquid from N.N.N. medium, which was then inoculated into the skin of the arm. No sores had developed after four months' observations. It is concluded that flies play no part in the carriage of the infection. These observers, however, have overlooked the possibility that flies may take up leishmania from a sore and pass them in their faeces on to an abrasion of the skin of another person within five minutes, as was shown by WENYON and O'CONNOR (1917) to occur in the case of Trichomonas.

Leishmaniasis of South America.—Direct observations on the means of transmission of the leishmaniasis of South America are few and far between, though there has been much speculation as to the possible vector. Biting flies, such as Tabanidae and Simulidae, as also ticks, have been suspected, but there is no experimental evidence to support these theories. TOWNSEND (1915), conducting investigations in Peru, suspected Chironomidae of the genus *Forcipomyia* as transmitting the disease. He found leishmania forms of a flagellate in the intestine of the flies, and by inoculating a guinea-pig in the skin with crushed flies, produced a papule in which he stated scanty leishmania forms occurred.

Now that Phlebotomus is generally regarded as the vector of oriental sore in the Old World, an American species, *P. lutzi*, is being suspected as playing a rôle in the spread of the American disease. So far there is no experimental evidence of this, but CERQUEIRA (1920) relates the experience of a professor of Bahia who developed a typical sore at the site of the puncture made by one of these flies. He states that four instances of a similar kind have come to light.

On the question of the relationship of the insect flagellates to *Leishmania tropica* and the culture of the flagellate, there is nothing to add to the remarks which were made in Part I of this review, save that the instance of the viability of the parasite in the cadaver referred to in connexion with the culture of *L. donovani* was an observation of PEDROSA (1917) of the successful culture of leishmania from a cutaneous ulcer in South America, 36 hours after death.

Prophylaxis.

Sterilization of all insect bites with iodine solution has been advocated as a means of avoiding infection, while on the assumption that the sand fly is the vector, protection from the ravages of these insects by means of sand fly nets (22 holes to the linear inch) is the natural procedure to adopt.

* From reliable information which the writer has received from India it appears that the structures observed by Mrs. ADIE were neither Leishman-Donovan bodies nor stages of any other flagellate, but some totally distinct organism.

* REFERENCES.

ACTON (1919), xiv, 205.
ADIE (1921), *Indian Jl. Med. Research*, ix, Oct. 255–260.
—— (192,2), *Indian Jl. Med. Research*, ix, No. 3 p. v.
DE AGUIAR PUPO (1921), *Bol. Soc. Med. e Cirurg. de S. Paulo*, iv, 97–99.
BANA (1920), xvii, 363.
BATES (1913), ii, 123.
BENOIT-GONIN (1911), *Bull. Soc. Path. Exot.*, iv, 182–184.
BLANC & CAMINOPETROS (1921), xvii, 361.
BONNE (1918), xiii, 101.
BOUILLIEZ (1917), x, 68.
—— (1918), xiii, 99.
CAMACHO (1914), Los progresso de la Clinica, Madrid, July.
CALICETI (1920), *L'Oto-Rhino-Laryngologie Internationale*, Dec. (see *Brit. Med. Jl.*, 1922, Jan. 21, Epitome of Current Medical Literature, p. 11).
CARINI (1911), *Bull. Soc. Path. Exot.*, iv, 289–291.
—— & PARANHOS (1909), *Bull. Soc. Path. Exot.*, ii, 255–257.
CASTELLANI (1913), i, 639.
CERQUEIRA (1919), xvii, 366.
CHALMERS & KAMAR (1920), xvi, 94.
CHATTON & BLANC (1918), xii, 156.
—— & —— (1918), xiv, 209.
CHRISTOPHERSON (1914), v, 274.
—— (1917), xi, 153.
CORNWALL & LA FRENAIS (1922), *Indian Jl. Med. Research*, ix, 533–547.
DUCKWORTH (1921), xviii, 241.
ESCOMEL (1916), ix, 236.
—— (1917), x, 70.
EVANS (1918), xiv, 206.
FLU (1911), *Centralb. f. Bakt.*, 1. Abt. Orig., lx, 624–637.
GACHET (1915), vii, 83.
GONDER (1913), ii, 449.
GRAY (1921), xvii, 364.
GREIG (1917), xii, 203.
HUNTEMÜLLER (1914), iii, 559.
INCHAUSTEGUI (1918), xvii, 366.
ITURBE (1917), x, 69.
LAVERAN (1917), "Leishmanioses," Paris, Masson et Cie.
—— & FRANCHINI (1921), xviii, 179.
LINDENBERG (1909), *Bull. Soc. Path. Exot.*, ii, 252–254.
LINDSAY (1914), iv, 402.
LOW (1915), vii, 84.
—— (1920), xvii, 362.
MACCORMAC (1919), xvi, 457.
MACKIE (1914), v, 262.
MARTOGLIO (1912), ii, 453.
MIGONE (1913), i, 639.
MITCHELL (1921), xvii, 364.
NATTAN-LARRIER, TOUIN & HECKENROTH (1909), *Bull. Soc. Path. Exot.*, ii, 587–591.
NELIGAN (1913), ii, 449.
NICOLLE, BLANC & LANGERON (1920), xvi, 456.
NICOLLE & MANCEAUX (1910), *C. R. Acad. Sci.*, cl, 889–891.
NICOLLE & SICRE (1908), *C. R. Soc. Biol.*, lxiv, 1096–1098.
ORMEROD (1920), xvii, 363.
PARROT (1919), xv, 442.
PATRICK (1920), xvi, 458.
PATTON (1919), xv, 442.
—— (1922), *Indian Jl. Med. Research*, ix, 496–532.
PEDROSA (1913), iii, 557.
VON PETERSEN (1914), v, 276.
PHOTINOS (1920), xvi, 457.
PRESSAT (1905), "La Paludisme et les Moustiques," Paris, Masson et Cie. [quoted by LAVERAN (1917)].
RAVAUT (1920), xvi, 457.
RODRÍGUEZ ARJONA (1915), vii, 86.
SCOTT (1917), xi, 154.
SEIDELIN (1912), i, 13.
SERGENT (Ed. & Et.) (1905), *C. R. Soc. Biol.*, lviii, 673–674.
SERGENT (Ed. & Et.), LEMAIRE & SÉNEVET (1915), vi, 227.
SERGENT (Ed. & Et.), PARROT (L.), DONATIEN (A.), & BÉGUET (M.) (1921), *C. R. Acad. Sci.*, clxxiii, 1030–1032.
DA SILVEIRA (1920), xvi, 458.
SINTON (1917), xi, 154.
—— (1921), xviii, 242.
STÉVENEL (1911), *Bull. Soc. Path. Exot.*, iv, 180–181.
STRONG & Others (1913), iii, 142.
SUSU (1917), xi, 153.
TEJERA (1920), xvi, 459.
THÉZÉ (1916), viii, 376.
TORRES (1920), xvi, 459; xvii, 365.
TOWNSEND (1915), viii, 12.
TRESTON (1921), xviii, 242.
WAGON (1914), iii, 558.
WENYON (1911), *Jl. London Sch. Trop. Med.*, i, 198.
—— & O'CONNOR (1917), *Jl. R.A.M.C.*, xxviii, No. 6, 686–698.
WERNER (H.) (1911), *Arch. f. Schiffs- u. Trop.-Hyg.*, xv, 581–585.
YAKIMOFF & SCHOKHOR (1914), iii, 556.

* References are to summaries in the *Tropical Diseases Bulletin* unless otherwise stated. Dates in parentheses refer to year of publication of original papers.

WELLCOME LIBRARY INSTITUTE